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* Please deliver to Examiner S. Devi in Art Unit 1645. *

Document(s) Transmitted: **Copy of proposed amended independent claims**

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In re. Patent Application of: Michael A. Apicella et al.
Serial No.: 09/077,572
Filed: October 13, 1998
Title: NON-TOXIC MUTANTS OF PATHOGENIC GRAM-NEGATIVE BACTERIA

Examiner: S. Devi
Group Art Unit: 1645
Docket No.: 875.001US2

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Docket No. 875.001US2
WD # 359462

UIRF # N5-50

NON-TOXIC MUTANTS OF PATHOGENIC GRAM-NEGATIVE BACTERIA
Applicant: Michael A. Apicella et al.
Serial No.: 09/077,572

22. [Amended] A method of making a mutant endotoxin comprising
mutating an *htrB* gene encoding a wild type endotoxin in [within] a wild type gram-negative bacterial pathogen to provide the mutant endotoxin; wherein the mutant endotoxin is the same as the wild type endotoxin except for [form an *htrB* mutant pathogen, wherein the *htrB* gene encodes an endotoxin] lacking one or more secondary acyl chains of lipid A [contained in a wild type gram-negative bacterial pathogen and lacking 3-hydroxy unsaturated C16 fatty acid substitutions on the lipid A as compared to a wild-type bacterial pathogen, and wherein the mutant endotoxin has substantially reduced toxicity when compared to the endotoxin of the wild type gram-negative bacterial pathogen, and
purifying the mutant endotoxin from the *htrB* mutant pathogen].

29. [Amended] A method for producing endotoxin-specific antisera, the method comprising
(a) immunizing an individual with a vaccine formulation comprising an *htrB* mutant of a gram-negative bacterial pathogen, endotoxin isolated from the *htrB* mutant of the gram-negative bacterial pathogen, or endotoxin purified from the *htrB* mutant of the gram-negative bacterial pathogen wherein the endotoxin is conjugated to a carrier protein; and
(b) collecting antibody produced from the immunized individual;
wherein the *htrB* mutant endotoxin is the same as wild type endotoxin except for lacking one or more secondary acyl chains of lipid A [lacks one or more secondary acyl chains of lipid A contained in a wild type gram-negative bacterial pathogen and lacks 3-hydroxy unsaturated C16 fatty acid substitutions on the lipid A as compared to a wild-type bacterial pathogen resulting in substantially reduced toxicity when compared to lipid A of the wild type gram-negative bacterial pathogen].